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(54) Title: NOVEL PEPTIDES USEFUL FOR INHIBITING BINDING OF LIPOPOLYSACCHARIDES (LPS) BY LIPOPOLYSACCHARIDE BINDING PROTEIN (LBP)

(57) Abstract

The present invention provides novel peptides derived from portions of the sequence of amino acids 95-104 of lipopolysaccharide binding protein (LBP). The invention also provides pharmaceutical compositions comprising the peptides of the invention, and diagnostic and therapeutic methods utilizing the peptides and pharmaceutical compositions of the invention.

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NOVEL PEPTIDES USEFUL FOR INHIBITING BINDING OF LIPOPOLYSACCHARIDES (LPS) BY LIPOPOLYSACCHARIDE BINDING PROTEIN (LBP)

Background of the Invention

This invention relates to peptides which inhibit binding of lipopolysaccharides (LPS) to lipopolysaccharide binding protein (LBP). Such peptides are foreseen to hold great promise as therapeutic applications for Gram-negative sepsis.

In Gram-negative sepsis, one of the most critical outcomes for the host is the activation of monocytic cells and the induction of cytokine production. While mild stimulation of the defense system via monocyte activation results in an appropriate beneficial response, excess production of cytokines or disturbance of the coagulation balance may be detrimental to the host.

Endotoxins are one of the biological mediators of Gram-negative sepsis syndrome. Endotoxins comprise a series of related LPS molecules that are present in the outer membrane of Gram-negative bacteria. The LPS molecules of smooth Gram-negative bacteria consist of a hydrophobic moiety, termed lipid A, a core oligosaccharide and a chain of repeating units of 1 to 7 sugars, termed the O chain (Rietschel *et al.*, *In Surface Structures of Microorganisms and their Interaction with the Mammalian Host*, **1988**, V.C.H., Frankfurt, FRG, p. 1). LPS molecules of rough Gram-negative bacteria consist of lipid A and a core oligosaccharide (Rietschel *et al.*, *In Surface Structures of Microorganisms*

and their Interaction with the Mammalian Host, **1988**, V.C.H., Franfurt, FRG, p. 1).

Different cells appear to be the targets of LPS and react in a way that subsequently leads to endotoxic shock. LPS causes B cells to proliferate and produce surface IgG (Dziarski, R., *Eur. J. Immunol.*, **1989**, *19*, 125). LPS also induces transcription of the HIV1 proviral genome in latently infected human monocytes (Pomerantz *et al.*, *J. Exp. Med.*, **1990**, *172*, 253). An important early event in LPS induced cell activation seems to be the tyrosine phosphorylation of a number of proteins in murine and human macrophages (Weinstein *et al.*, *Proc. Natl. Acad. Sci. USA*, **1991**, *88*, 4148).

LPS stimulates macrophages to release mediators including cytokines, such as interleukin-1 (IL-1) (Cavaillon, J.M. and N. Haeffner-Cavaillon, *Cytokine*, **1990**, *2*, 1), IL-6 (Fong *et al.*, *J. Immunol.*, **1989**, *142*, 2321), IL-8 (Yoshimura *et al.*, *J. Immunol.*, **1987**, *139*, 788) and tumor necrosis factor (TNF) (Beutler, B. and A. Cerami, *Nature*, **1986**, *320*, 584), lipid products derived from arachidonic acid, hydrolytic enzymes and toxic oxygen radicals (Nathan, C.F., *J. Clin. Invest.*, **1987**, *79*, 319). Among the targets of these substances are phagocytic cells that are activated as a part of the host defense against infection. However, these same mediators may also cause injury to host cells resulting in changes that lead to septic shock, multiorgan failure, respiratory distress syndrome, and death (Ziegler *et al.*, *N. Eng. J. Med.*, **1991**, *324*, 429; Danner *et al.*, *Chest*, **1991**, *99*, 169). LPS acts at picomolar concentrations making it one of the most potent macrophage agonists known.

Most of the circulating LPS in serum is bound to a protein termed LBP (Tobias *et al.*, *J. Exp. Med.*, **1986**, *164*, 777). LBP cDNA has been cloned and sequenced (Schumann *et al.*, *Science*, **1990**, *249*, 1429). LBP is produced in hepatocytes as a 50 kDA protein and is constitutively secreted into the bloodstream as a 60 kDA glycoprotein at a concentration of approximately 500 ng/ml (Ulevitch *et al.*, *In Endotoxins, from Pathophysiology to Therapeutic Approaches*,

1990, Medicine-Sciences Flammarian, Paris, p. 31). The protein concentration rises in the acute phase to 50 μg/ml, and LBP, which does not have activity by itself, binds to LPS with high affinity. Upon binding to the lipid A moiety of LPS (Mathison *et al.*, *J. Immunol.*, **1992**, *149*, 200; Tobias *et al.*, *J. Biol. Chem.*, **1989**, *264*, 10867), LBP does not suppress or block the effects of LPS, but enhances endotoxin effects. LPS-induced TNF production and TNF mRNA expression in rabbit peritoneal macrophages is strongly enhanced when LPS is complexed to LBP (Schumann *et al.*, *Science*, **1990**, *249*, 1429). Macrophages detect and bind LPS much more rapidly when it is complexed with LBP (Schumann *et al.*, *Science*, **1990**, *249*, 1429). Binding of LBP to LPS on the surface of bacteria or to LPS inserted into erythrocyte membranes dramatically enhances their interaction with macrophages (Wright *et al.*, *J. Exp. Med.*, **1989**, *170*, 1231). Thus LBP acts as an opsonin for Gram-negative bacteria (Wright *et al.*, *J. Exp. Med.*, **1989**, *170*, 1231).

Several methodologic approaches have been used to identify receptors on monocytes for LBP/LPS complexes including studies using radiolabelled LPS (Haeffner-Cavaillon *et al.*, *J. Immunol.*, **1982**, *128*, 1950; Tobias *et al.*, *J. Immunol.*, **1993**, *150*, 3011), isolation of the molecules by cross-linking of LPS to monocyte membranes (Lei, M.G. and D.C. Morrison, *J. Immunol.*, **1988**, *141*, 996), and blocking of cellular functions with anti-monocyte antibodies (Wright, S.D. and M.T.C. Jong, *J. Exp. Med.*, **1986**, *164*, 1876; Wright *et al.*, *Science*, **1990**, *249*, 1431). Evidence from several groups indicates that at least the four following molecules or complexes serve as binding sites and/or receptors for LPS on human monocytes: the CD11/CD18 Leu-CAM complex (Wright, S.D. and M.T.C. Jong, *J. Exp. Med.*, **1986**, *164*, 1876), an 80 kDa molecule of mouse and human leukocytes (Lei, M.G. and D.C. Morrison, *J. Immunol.*, **1988**, *141*, 996; Lei, M.G. and D.C. Morrison, *J. Immunol.*, **1988**, *141*, 1006), a lectin-like monocyte membrane molecule interacting with the polysaccharide moiety of LPS (Haeffner-Cavaillon *et al.*,

Cell. Immunol., **1985**, *91*, 119) and the glycolipid-anchored CD14 molecule (Wright *et al.*, *Science*, **1990**, *249*, 1431).

The molecule on the cell surface that mediates the binding of LBP/LPS complexes is restricted to monocytes, neutrophils, and macrophages, is mobile in the plane of the membrane and is distinct from receptors for other known opsonins (Wright *et al.*, *J. Exp. Med.*, **1989**, *170*, 1231). CD14 is a 53 kDa glycoprotein found on the surface of myeloid cells (Goyert *et al.*, *J. Immunol.*, **1986**, *137*, 3909). CD14 is linked to the cell surface via a phosphatidylinositol (PI) anchor (Haziot *et al.*, *J. Immunol.*, **1988**, *141*, 547). A soluble form of the protein also exists and can be found in human serum (Bazil *et al.*, *Mol. Immunol.*, **1989**, *26*, 657). CD14 is located on chromosome 5 in a region known to encode for several cytokines and loss of this region (q23-31) is associated with certain forms of leukemia (Goyert *et al.*, *Science*, **1988**, *239*, 497). Complexes of LPS and LBP are recognized and specifically bound by CD14 on myeloid cells (Wright *et al.*, *Science*, **1990**, *249*, 1431; Couturier *et al.*, *J. Immunol.*, **1991**, *147*, 1899). Antibody to CD14 has been shown to inhibit LBP/LPS-mediated effects, including TNF production and opsonin function (Wright *et al.*, *Science*, **1990**, *249*, 1431). Antibody to CD14 has also been shown to block LPS-induced tyrosine phosphorylation (Weinstein *et al.*, *J. Leuk. Biol.*, **1991**).

A method currently under clinical investigation for interfering with processes that lead to endotoxic shock involves the application of anti-LPS antibodies to patients in shock (Ziegler *et al.*, *N. Eng. J. Med.*, **1991**, *324*, 429). However, it is clearly beneficial to develop methods of therapy which inhibit the development of toxic shock as opposed to methods of treating patients who already have developed toxic shock or other manifestations of Gram-negative sepsis due to endotoxin.

Summary of the Invention

Recently, we have cloned the gene for LBP and have expressed it as an immunoglobulin fusion protein (LBPfp). We have also constructed a series of nested LBP peptides and found, unexpectedly, that two of these peptides, comprising amino acids 91-105 and 94-108 of the mature protein, specifically inhibit the binding of LPS to Fc-captured LBPfp. Our further work has suggested that a series of peptides having as their core region portions of the 95-104 amino acid sequences of the LBP should be extremely useful for therapeutic applications in the prevention and treatment of Gram-negative sepsis.

This invention therefore relates to novel peptides having the Formula:

R^1-X-A-B-C-D-E-F-G-H-I-J-Y-R^2 wherein:

A is selected from the group consisting of D- or L-lysine, D- or L-arginine, D- or L-alanine, and D- or L-histidine;

B is selected from the group consisting of D- or L-serine, D- or L-threonine, D- or L-alanine, and D- or L-arginine;

C is selected from the group consisting of D- or L-phenylalanine, D- or L-alanine, D- or L-tyrosine, D- or L-serine, and D- or L-proline;

D is selected from the group consisting of D- or L-phenylalanine, D- or L-tyrosine, D- or L-leucine, and D- or L-alanine;

E is selected from the group consisting of D- or L-lysine, D- or L-arginine, D- or L-alanine, and D- or L-histidine;

F is selected from the group consisting of D- or L-leucine, D- or L-alanine, and D- or L-methionine;

G is selected from the group consisting of D- or L-glutamine, D- or L-asparagine, D- or L-lysine, D- or L-serine, and D- or L-alanine;

H is selected from the group consisting of D- or L-glycine, and D- or L-alanine;

I is selected from the group consisting of D- or L-serine, D- or L-threonine, D- or L-asparagine, and D- or L-alanine;

J is selected from the group consisting of D- or L-phenylalanine, D- or L-alanine, and D- or L-tyrosine;

X is a linear chain of from zero to four amino acids;

Y is a linear chain of from zero to five amino acids;

R^1 is H (signifying a free-terminal amino group), formyl, lower alkyl, aryl, lower alkanoyl, aroyl, biotinyl, alkyloxycarbonyl, aryloxycarbonyl or desamino (signifying no alpha amino group on the N-terminal amino acid); and

R^2 is H (signifying descarboxy where the α carboxyl group in the C-terminal amino acid is absent) or OR^3, where R^3 is H (signifying a free carboxylic acid on a C-terminal carboxyl group) or lower alkyl or aryl, NR^4R^5 where R^4 and R^5 are each selected independently from H, lower alkyl, aryl, or taken together are a methylene chain of 4-8 methylene groups ($-(CH_2)_n-$ where n = 4 to 8);

and pharmaceutically acceptable salts thereof.

Peptides of the Formula have as their core region portions of the 95-104 amino acid sequences of the LBP, with residue 1 defined as the N-terminus of the mature proteins after the cleavage of the signal peptides.

Tests indicate that peptides of the Formula inhibit the binding of LPS in concentrations of peptide ranging from about 1 to about 1500 μm. Tests also indicate that certain alterations within the core sequence, including the addition or deletion of amino acids, do not result in loss of biological activity.

This invention relates not only to the novel peptides of the Formula, but also to pharmaceutical compositions comprising them, to diagnostic and therapeutic methods utilizing them, and to methods of preparing them.

Brief Description of th Drawings

Figure 1 is a graph showing the ability of different concentrations of LBPfp to bind different concentrations of LPS-HRP.

Figure 2 is a graph showing the competitive inhibition of LPS-HRP binding to Fc-captured LBPfp. The dashed lines indicate the IC50 values for the competitors. Vertical bars indicate the standard error of the mean.

Figure 3 is a graph showing the IC50 values for the peptides of Examples 1-8 in the ELISA competitive assay.

Figure 4 provides graphs showing the results of LAL experiments. The dotted lines indicate the IC50 values for each of the inhibitors. Figure 4a compares the neutralization potency of the peptides of Examples 1, 4 and 5 and polymyxin B in a 0111LPS (smooth challenge). Figure 4b shows the ability of the peptide of Example 1 and polymyxin B to block the LAL reaction to 1 pg/ml rough LPS. Figure 4c shows the ability of the three tested peptides to inhibit the LAL reaction induced by 500 pg/ml lipid A.

Figure 5 is a graph showing the blocking of PBMC response to LPS challenge by the peptide of Example 1. The amount of TNF found in cultures (vertical axis) stimulated with increasing concentrations of LPS (horizonal axis), an index of monocyte activation by LPS, is shown. The different curves represent LPS dose responses in the presence of different amounts of the peptide. The dotted lines show the amount of LPS each peptide concentration tested can inhibit by 50%.

Figure 6 is a graph showing the ability of the peptide of Example 6 to inhibit PBMC response to 1% normal human serum-potentiated LPS challenge. Shown is the ability of different concentrations of the peptide (horizontal axis) to inhibit 1ng/mL and 10 ng/mL 0111 LPS challenge in the constant presence of 1% normal human serum. The dotted lines indicate the IC50 of the peptide when tested with 1ng/mL and 10 ng/mL LPS.

Detailed Description of the Invention

Preferred peptides of this invention are those of the Formula as previously defined, wherein X is selected from the following group (or analogs thereof wherein one or more of the amino acids are replaced with the analogous D-amino acid):

Trp-Lys-Val-Arg
Lys-Val-Arg
Val-Arg
Arg
Gln
Lys
Ala
His
and null (signifying no amino acid)

Also preferred are peptides having the Formula as previously defined, wherein Y is selected from the following group (or analogs thereof wherein one or more of the amino acids are replaced with the analogous D-amino acid): Asp
Asp-Val-Ser-Val
Asp-Val
Asp-Val-Ser
Glu-Val-Ser-Val
Ala-Val-Ser-Val
His-Val-Ser-Val
Lys-Val-Ser-Val
Arg-Val-Ser-Val
Asp-Val-Ser-Val
Asp-Leu-Ser-Val
Asp-Lys-Ser-Val
Asp-Val-Ala-Val
Asp-Val-Thr-Val
Asp-Val-Tyr-Val
Asp-Val-Ser-Val
Asp-Val-Ser-Ala
Asp-Val-Ser-Ile

and null (signifying no amino acid).

Representative examples of specifically preferred peptides include the following "Preferred Peptides" (SEQ ID NOS:1-64):

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 1)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 2)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2 (SEQ ID NO: 3)

Acetyl-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 4)

Acetyl-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 5)

D-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2

D-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2

D-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2

Gln-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 6)

Lys-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 7)

Ala-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 8)

His-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 9)

Arg-Arg-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 10)

Arg-Ala-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 11)

Arg-His-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 12)

Arg-Lys-Thr-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 13)

Arg-Lys-Ala-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 14)

Arg-Lys-Arg-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 15)

Arg-Lys-Ser-Ala-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 16)

Arg-Lys-Ser-Tyr-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 17)

Arg-Lys-Ser-Ser-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 18)

Arg-Lys-Ser-Pro-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 19)

Arg-Lys-Ser-Phe-Tyr-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 20)

Arg-Lys-Ser-Phe-Leu-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 21)

Arg-Lys-Ser-Phe-Ala-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 22)

Arg-Lys-Ser-Phe-Phe-Arg-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 23)

Arg-Lys-Ser-Phe-Phe-Ala-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 24)

Arg-Lys-Ser-Phe-Phe-His-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 25)

Arg-Lys-Ser-Phe-Phe-Lys-Ala-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 26)

Arg-Lys-Ser-Phe-Phe-Lys-Met-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 27)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Asn-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 28)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Lys-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 29)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Ser-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 30)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Ala-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 31)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Ala-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 32)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Thr-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 33)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Asn-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 34)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ala-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 35)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Tyr-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 36)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Ala-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 37)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Glu-Val-Ser-Val-NH_2 (SEQ ID NO: 38)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Ala-Val-Ser-Val-NH_2 (SEQ ID NO: 39)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-His-Val-Ser-Val-NH_2 (SEQ ID NO: 40)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Lys-Val-Ser-Val-NH_2 (SEQ ID NO: 41)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Arg-Val-Ser-Val-NH_2 (SEQ ID NO: 42)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Ala-Ser-Val-NH_2 (SEQ ID NO: 43)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Leu-Ser-Val-NH_2 (SEQ ID NO: 44)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Lys-Ser-Val-NH_2 (SEQ ID NO: 45)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ala-Val-NH_2 (SEQ ID NO: 46)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Thr-Val-NH_2 (SEQ ID NO: 47)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Tyr-Val-NH_2 (SEQ ID NO: 48)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Tyr-NH_2 (SEQ ID NO: 49)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Ala-NH_2 (SEQ ID NO: 50)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Ile-NH_2 (SEQ ID NO: 51)

Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 52)

Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 53)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 54)

Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 55)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-NH_2 (SEQ ID NO: 56)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-NH_2 (SEQ ID NO: 57)

~~Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2 (SEQ ID NO: 58)~~

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-NH_2 (SEQ ID NO: 59)

Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-NH_2 (SEQ ID NO: 60)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-NH_2 (SEQ ID NO: 61)

Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2 (SEQ ID NO: 62)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2 (SEQ ID NO: 63)

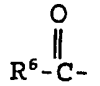
Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2 (SEQ ID NO: 64)

D-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-Lys(Biotin)-NH_2

D-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2

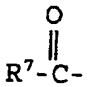
As used herein, the term "alkyl" includes branched, straight-chain, and cyclic saturated hydrocarbons. The term "lower alkyl" means an alkyl having from one to six carbon

atoms, such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, t-butyl, pentyl, isopentyl, neopentyl, cyclopentylmethyl and hexyl. The term "alkanoyl" means



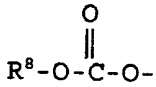
wherein R^6 is a alkyl group.

The term "aroyl" means



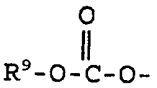
wherein R^7 is an aryl group. The term "aryl" means an aromatic or heteroaromatic structure having between one and three rings, which may or may not be ring fused structures, and are optionally substituted with halogens, carbons, or other heteroatoms such as nitrogen (N), sulfur (S), phosphorus (P), and boron (B).

The term alkoxycarbonyl means



wherein R^8 is a lower alkyl group.

The term aryloxycarbonyl means



wherein R^9 is an aryl and arylmethyl group.

Halogen refers to fluorine, chlorine, bromine or iodine.

The term "terminal α-amino group of X" refers to the α-amino group of the N-terminal amino acid of X.

The peptides of the Formula can be used in the form of the free peptide or a pharmaceutically acceptable salt. Amine salts can be prepared by treating the peptide with an acid according to known methods. Suitable acids include inorganic acids such as hydrochloric acid, hydrobromic acid, perchloric acid, nitric acid, thiocyanic acid, sulfuric acid, and phosphoric acid, and organic acids such as formic acid, acetic acid, propionic acid, glycolic acid, lactic acid,

pyruvic acid, oxalic acid, malonic acid, succinic acid, maleic acid, fumaric acid, anthranilic acid, cinnamic acid, naphthalenesulfonic acid, and sulfanilic acid.

Carboxylic acid groups in the peptide can be converted to a salt by treating the peptide with a base according to known methods. Suitable bases include inorganic bases such as sodium hydroxide, ammonium hydroxide, and potassium hydroxide, and organic bases such as mono-, di-, and tri-alkyl and aryl amines (e.g., triethylamine, diisopropylamine, methylamine, and dimethylamine and optionally substituted mono-, di, and tri-ethanolamines.

As referred to herein, the amino acid components of the peptides and certain materials used in their preparation are identified by abbreviations for convenience. These abbreviations are as follows:

Amino Acid	Abbreviations	
L-alanine	Ala	A
D-alanine	D-Ala	a
L-arginine	Arg	R
D-arginine	D-Arg	r
D-asparagine	D-Asn	N
L-asparagine	Asn	n
L-aspartic acid	Asp	D
D-aspartic acid	D-Asp	d
L-cysteine	Cys	C
D-cysteine	D-Cys	c
L-glutamic acid	Glu	E
D-glutamic acid	D-Glu	e
L-glutamine	Gln	Q
D-glutamine	D-Gln	q
glycine	Gly	G
L-histidine	His	H
D-histidine	D-His	h
L-isolelucine	Ile	I
D-isolelucine	D-Ile	i
L-leucine	Leu	L
D-leucine	D-Leu	l
L-lysine	Lys	K
D-lysine	D-Lys	k
L-phenylalanine	Phe	F
D-phenylalanine	D-Phe	f
L-proline	Pro	P
D-proline	D-Pro	p
L-pyroglutamic acid	pGlu	
D-pyroglutamic acid	DpGlu	
L-serine	Ser	S
D-serine	D-Ser	s
L-threonine	Thr	T
D-threonine	D-Thr	t
L-tyrosine	Tyr	Y
D-tyrosine	D-Tyr	y
L-tryptophan	Trp	W
D-tryptophan	D-Trp	w
L-valine	Val	V
D-valine	D-Val	v
L-alloisolucine	Allo	
D-alloisolucine	D-Allo	
L-methionine	Met	M
D-methionine	D-Met	m

Reagents	Abbreviations
Trifluoroacetic acid	TFA
Methylene chloride	CH_2Cl_2
N,N-Diisopropylethylamine	DIEA
N-Methylpyrrolidone	NMP
1-Hydroxybenzotriazole	HOBT
Dimethylsulfoxide	DMSO
Acetic anhydride	Ac_2O
Diisopropylcarbodiimide	DIC

Amino acids preceded by L- or D- refer, respectively, to the L- or D- enantiomer of the amino acid, whereas amino acids not preceded by L- or D- refer to the L-enantiomer. Common derivatives of any of the amino acids may also be incorporated into the peptides of this invention (e.g., Lys(Biotin) for Lys).

Methods of Preparation of Peptides

The peptides can generally be prepared following known techniques, as described, for example, in the cited publications, the teachings of which are specifically incorporated herein. In a preferred method, the peptides are prepared following the solid-phase synthetic technique initially described by Merrifield in J.Amer.Chem.Soc., 85, 2149-2154 (1963). Other techniques may be found, for example, in M. Bodanszky, et al, Peptide Synthesis, second edition, (John Wiley & Sons, 1976), as well as in other reference works known to those skilled in the art.

Appropriate protective groups usable in such syntheses and their abbreviations will be found in the above text, as well as in J.F.W. McOmie, Protective Groups in Organic Chemistry, (Plenum Press, New York, 1973). The common protective groups used herein are t-butyloxycarbonyl (Boc), fluorenylmethoxycarboyl (FMOC), benzyl (Bzl), tosyl (Tos), o-bromo-phenylmethoxycarbonyl (BrCBZ), phenylmethoxycarbonyl (CBZ), 2-chloro-phenylmethoxycarbonyl (2-Cl-CBZ), 4-methoxy-2,3,6-trimethylbenzenesulfonyl (Mtr), trityl (Trt), formyl (CHO), and tertiary butyl (t-Bu).

General synthetic procedures for the synthesis of peptides of the Formula by solid phase methodology are as follows:

A. General Synthetic Procedures For Solid Phase Peptide Synthesis Using N^{α}-Boc Protection

		REPETITIONS	TIME
1.	25% TFA in CH_2Cl_2	1	3 min.
2.	50% TFA in CH_2Cl_2	1	16 min.
3.	CH_2Cl_2	5	3 min.
4.	5% DIEA in NMP	2	4 min.
5.	NMP	6	5 min.
6.	Coupling step	1	57 min.
	a. Preformed BOC-Amino Acid-HOBT active ester in NMP		37 min.
	b. DMSO		16 min.
	c. DIEA		5 min.
7.	10% Ac_2O, 5% DIEA in NMP	1	9 min.
8.	CH_2Cl_2	5	3 min.

B. General Synthetic Procedure For Solid Phase Peptide Synthesis Using N^{α}-FMOC Protection

		REPETITIONS	TIME
1.	50% piperidine in NMP	1	1 min.
2.	50% piperidine in NMP	1	12 min.
3.	NMP wash	7	1 min.
4.	Coupling	2	30 min.
	FMOC amino acid dissolved in HOBT/NMP followed by the addition of DIC (di-isopropylcarbodiamide) in NMP.		
5.	NMP wash.	2	1 min.

In the solid phase synthesis approach the C-terminal amino acid is attached to the appropriately functionalized resin. This can be a resin such as 4-methylbenzhydrylamine-polystyrene-divinylbenzene resin where the C-terminal amide is desired, BOC protection of the alpha amino groups is desired and HF cleavage of the peptide from the resin is desired or HMP-resin (4-hydroxymethylphenoxymethyl-styrene-divinylbenzene resin) where the C-terminal acid is desired, FMOC protection of the alpha amino group is desired and TNF

cleavage of the peptide from the resin is desired. The selection of the appropriate resin, protection strategy and cleavage strategy is done according to procedures by those skilled in the art of peptide synthesis. Following attachment of the appropriately protected C-terminal amino acid to the resin, the alpha amino protecting group is removed to expose the free alpha amino group. The desired protected, partially protected or unprotected amino acid is then coupled to the free amino group using reagents or techniques that will form the desired amide bond. This can involve preactivation of the carboxyl and/ or amino groups or their conversion into chemical moieties which, upon reaction, will give the desired amides. Such techniques involve the use of carbodiimides, formation of active esters, active anhydrides or other such procedures employed in the synthesis of peptides. Examples of such techniques can be found in "The Peptides, Analysis, Synthesis, Biology", Volume 1, Major Methods of Peptide Bond Formation, Eds. E. Gross and J. Meienhofer, Academic Press, NY, 1979. As an alternative to the stepwise addition of amino acids to the peptide resin, preformed blocks of two or more amino acids may be added. The sequence of deprotection and coupling is repeated until assembly of the desired peptide on the resin is complete. The protecting groups are removed from the peptide and the peptide removed from the resin. These two steps can be accomplished either consecutively or concurrently. The peptide is then isolated from the resin and purified to an appropriate degree of purity.

In a solution phase approach for preparing peptides, an appropriately protected amino acid is coupled to a second appropriately protected amino acid by forming an amide bond between them. Such techniques involve the use of carbodiimides, formation of active esters, active anhydrides or other such procedures employed in the synthesis of peptides. The amine bond may also be formed through the use of enzymes, functioning in an manner described as reverse proteolysis. Following appropriate deprotection to generate

free amino groups or carboxylic acid groups, additional appropriately protected amino acids or preformed blocks of amino acids are covalently linked through the formation of amide bonds. The selection and removal of appropriate functional group protection is according to procedures known to those skilled in the art of peptide chemistry. Examples of such techniques can be found in "The Peptides, Analysis, Sysnthesis, Biology", Volume 3, Protection of Functional Groups in Peptide Synthesis, Eds. E. Gross and J. Meienhofer, Academic Press, NY, 1981. Once assembly of the desired peptide is complete, functional group protection is removed and the peptide purified to the desired degree of homogeneity. Examples 1 through 8 were prepared using the solid phase strategy with BOC protection and HF cleavage from the resin. The peptides in Figures 2 and 3 were prepared using the solid phase strategy with FMOC protection and TFA cleavage from the resin.

The peptides can also be prepared using standard genetic engineering techniques known to those skilled in the art. For example, the peptide can be produced enzymatically by inserting nucleic acid encoding the peptide into an expression vector, expressing the DNA, translating the DNA into RNA and the RNA into the peptide. The peptide is then purified using chromatographic or electrophoretic techniques, or by means of a carrier protein which can be fused to, and subsequently cleaved from, the peptide by inserting into the expression vector in phase with the peptide encoding sequence a nucleic acid sequence encoding the carrier protein. The fusion protein-peptide may be isolated using chromatographic, electrophoretic or immunological techniques (such as binding to a resin via an antibody to the carrier protein). The peptide can be cleaved using chemical methodology or enzymatically, as by, for example, hydrolases.

Methods of Preparation of Pharmaceutical Compositions

Pharmaceutical compositions of this invention comprise a pharmaceutically acceptable carrier or diluent and an

effective quantity of one or more of the peptides of the Formula or an acid or base salt thereof. The carrier or diluent may take a wide variety of forms depending on the form of preparation desired for administration, e.g., sublingual, rectal, nasal, oral, transdermal or parenteral.

In preparing the compositions in oral dosage form, any of the usual pharmaceutical media may be employed, for example, waters, oils, alcohols, flavoring agents, preservatives, and coloring agents, to make an oral liquid preparation (e.g., suspension, elixir, or solution) or with carriers such as starches, sugars, diluents, granulating agents, lubricants, binders, and disintegrating agents, to make an oral solid preparation (e.g., powder, capsule, or tablet).

Controlled release forms or enhancers to increase bioavailability may also be used. Because of their ease in administration, tablets and capsules represent the most advantageous oral dosage unit form, in which case solid pharmaceutical carriers are employed. If desired, tablets may be sugar coated or enteric coated by standard techniques.

For parenteral products, the carrier will usually be sterile water, although other ingredients to aid solubility or as preservatives may be included. Injectable suspensions may also be prepared, in which case appropriate liquid carriers and suspending agents can be employed.

The peptides can also be administered locally at a wound or inflammatory site by topical application of a solution or cream.

Alternatively, the peptide may be administered in liposomes or microspheres (or microparticles). Methods for preparing liposomes and microspheres for administration to a patient are known to those skilled in the art. U.S. Patent No. 4,789,734 describes methods for encapsulating biological materials in liposomes. Essentially, the material is dissolved in an aqueous solution, the appropriate phospholipids and lipids added, along with surfactants if required, and the material dialyzed or sonicated, as

necessary. A review of known methods is by G. Gregoriadis, Chapter 14, "Liposomes", Drug Carriers in Biology and Medicine, pp. 287-341 (Academic Press, 1979). Microspheres formed of polymers or proteins are well known to those skilled in the art, and can be tailored for passage through the gastrointestinal tract directly into the bloodstream. Alternatively, the peptide can be incorporated and the microspheres, or composite of microspheres, implanted for slow release over a period of time, ranging from days to months. See, for example, U.S. Patents Nos. 4,906,474, 4,925,673 and 3,625,214.

The peptides are generally active when administered parenterally in amounts above about 1 μg/kg body weight. Effective doses by other routes of administration are generally those which result in similar blood level to i.v. doses above about 10 μg/Kg. For treatment to prevent organ injury in cases involving reperfusion, the peptides may be administered parenterally in amounts from about 0.01 to about 10 mg/kg body weight. Generally, the same range of dosage amounts may be used in treatment of other diseases or of conditions where inflammation is to be reduced. This dosage will be dependent, in part, on whether one or more peptides are administered.

Methods for Demonstrating Binding

Peptides that are biologically active are those which inhibit binding of LPS to LBP.

Peptides can be screened for their ability to inhibit such binding, for example, using ELISA-based assay to detect lipid A binding determinants. Such an assay utilizes a fusion protein, immobilized onto EIA plates, comprised of human LBP fused to human IgG constant domains (LPSfp). Horseradish peroxidase-labelled LPS (HLPS-HRP) bound by the LBP moiety of the LBPfp fusion protein is added, and subsequently is detected by the addition of a chromogenic substrate. However, since LBP is specific for the lipid A portion of LPS molecules, if a Lipid A-binding test substance

is added with the labelled-LPS, it would be expected to inhibit binding of the labelled-LPS to the LBPfp. Thus, this screening assay could be used to identify test substances that can competitively inhibit binding of LPS to LBPfp.

Clinical Applications

Since the peptides of this invention inhibit binding of LPS to LBP, they should be useful for the prevention or treatment of Gram negative septic shock.

The criteria for assessing response to therapeutic modalities employing these peptides, and, hence, effective dosages of the peptides of this invention for treatment, are dictated by the specific condition and will generally follow standard medical practices. For example, the criteria for the effective dosage to prevent LPS induced toxicity would be determined by one skilled in the art by looking at cytokine release and by monitoring clinical response.

Diagnostic Reagents

The peptides can also be used for the detection of human disorders in which LPS is implicated. Such elevated LPS levels would, for example, be seen in patients with sepsis. Patient samples can be collected and tested using the peptide to bind to LPS and the resulting peptide-LPS or peptide-LPS-CD14 conjugate detected or quantitated using direct or indirect procedures.

The peptide is labeled radioactively, with a fluorescent tag, enzymatically, or with electron dense material such as gold for electron microscopy. The LPS can also be measured with ELISA or radioimmunoassay procedures, using labeled peptide as the trapping or the detecting reagent.

The following examples are presented to illustrate, not limit, the invention. In the examples and throughout the specification, parts are by weight unless otherwise indicated.

EXAMPLE 1: Preparation of arginyl-lysyl-seryl-ph nylalanyl-ph nylalanyl-lysyl-leucyl-glutamyl-glycyl-seryl-phenylalanyl-aspartyl-valyl-seryl-valine-amide (SEQ ID NO: 1)

The peptide was prepared on an ABI Model 431A Peptide Synthesizer using Version 1.12 of the standard BOC software. 4-methyl benzhydrylamine resin (0.46 g, 0.5 mmol) was used in the synthesis. The final weight of the resin was 1.82 g.

The peptide was cleaved from the resin (1.74 g) using 18 mL of HF and 1.75 mL of anisole for 60 min at 0°C. The resin was washed with ether and the peptide extracted with a 1:1 solution of TFA:CH_2Cl_2 to give 964 mg of crude peptide.

The crude peptide (964 mg) was purified on a Vydac C-18 column (15μ, 5 x 25 cm) eluting with a 25-75% gradient of 80% ethanol in 0.1% TFA over 120 min at a flow rate of 15 mL per min. Fractions were collected, analyzed by HPLC and pure fractions pooled and lyophilized to give 178 mg of semipure peptide.

178 mg of semipure peptide was purified using a Vydac C-18 column (15μ, 5 x 25 cm) eluting with a 15-65% gradient of 80% acetonitrile in 0.1% TFA over 120 min at a flow rate of 15 mL per min. Fractions were collected, analyzed by HPLC, pooled and lyophilized to give 126 mg of pure peptide.

Amino acid analysis: Arg 0.98 (1), Asx 1.05 (1), Glx 0.99 (1), Gly 1.02 (1), Leu 1.03 (1), Lys 1.99 (2), Phe 2.98 (3), Ser 1.82 (3), Val 2.01 (2).

FAB/MS: MH^+ 1744.3

EXAMPLE 2: Preparation of tryptophyl-lysyl-valyl-arginyl-lysyl-seryl-phenylalanyl-phenylalanyl-lysyl-leucyl-glutamyl-glycyl-seryl-phenylalanyl-aspartyl-valyl-seryl-valine-amide (SEQ ID NO: 2)

The peptide was prepared on an ABI Model 431A Peptide Synthesizer using Version 1.12 of the standard BOC software. 4-methyl benzhydrylamine resin (0.46 g, 0.5 mmol) was used in the synthesis. The final weight of the resin was 1.97 g.

The peptide was cleaved from the resin (1.84 g) using 19 mL of HF and 1.85 mL of anisole for 60 min at 0°C. The resin was washed with ether and the peptide extracted with a 1:1 solution of TFA:CH_2Cl_2 to give 858 mg of crude peptide.

The crude peptide (858 mg) was purified on a Vydac C-18 column (15μ, 10 x 30 cm) eluting with a 20-50% gradient of 80% acetonitrile in 0.1% TFA over 120 min at a flow rate of 60 mL per min. Fractions were collected, analyzed by HPLC and pure fractions pooled and lyophilized to give 105 mg.

Amino acid analysis: Arg 1.0 (1), Asx 1.58 (1), Glx 1.34 (1), Gly 1.09 (1), Leu 1.08 (1), Lys 2.41 (3), Phe 3.0 (3), Ser 1.79 (3), Trp 0.94 (1), Val 3.08 (3).

FAB/MS: MH^+ 2158.1

EXAMPLE 3: Preparation of Tryptophyl-lysyl-valyl-arginyl-lysyl-seryl-phenylalanyl-phenylalanyl-lysyl-leucyl-glutamyl-glycyl-seryl-phenylalanyl-aspartic acid-amide (SEQ ID NO: 3)

The peptide was prepared on an ABI Model 431A Peptide Synthesizer using Version 1.12 of the standard BOC software. 4-methyl benzhydrylamine resin (0.46 g, 0.5 mmol) was used in the synthesis. The final weight of the resin was 1.84 g.

The peptide was cleaved from the resin (1.76 g) using 18 mL of HF and 1.8 mL of anisole for 60 min at 0°C. The resin was washed with ether and the peptide extracted with a 1:1 solution of TFA:CH_2Cl_2 to give 1.01 g of crude peptide.

The crude peptide (1.01 g) was purified on a Vydac C-18 column (15μ, 5 x 25 cm) eluting with a 25-65% gradient of 80% ethanol in 0.1% TFA over 120 min at a flow rate of 15 mL per min. Fractions were collected, analyzed by HPLC and pure fractions pooled and lyophilized to give 251 mg.

Amino acid analysis: Arg 0.94, (1), Asx 1.45 (1), Glx 1.17 (1), Gly 1.03 (1), Leu 1.05 (1), Lys 2.87 (3), Phe 2.98 (3), Ser 1.36 (2), Trp 0.83 (1), Val 0.95 (1).

FAB/MS: MH^+ 1872.3

EXAMPLE 4: Preparation of acetyl-arginyl-lysyl-seryl-phenylalanyl-phenylalanyl-lysyl-leucyl-glutamyl-glycyl-seryl-phenylalanyl-aspartyl-valyl-seryl-valine-amide (SEQ ID NO: 4)

The peptide was prepared on an ABI Model 431A Peptide Synthesizer using Version 1.12 of the standard BOC software. 4-methyl benzhydrylamine resin (0.58 g, 0.5 mmol) was used in the synthesis. The final weight of the resin was 2.21 g.

The peptide was cleaved from the resin (2.21 g) using 22 mL of HF and 2.2 mL of anisole for 60 min at 0°C. The resin was washed with ether and the peptide extracted with a 1:1 solution of TFA:CH_2Cl_2 to give 1.31 g of crude peptide.

The crude peptide (1.00 g) was purified on a Vydac C-18 column (15μ, 5 x 25 cm) eluting with a 35-65% gradient of 80% ethanol in 0.1% TFA over 120 min at a flow rate of 15 mL per min. Fractions were collected, analyzed by HPLC and pure fractions pooled and lyophilized to give 132 mg of semipure peptide.

Semipure peptide (132 mg) was purified on a Vydac C-18 column (15μ, 5 x 25 cm) eluting with a 25-50% gradient of 80% acetonitrile in 0.1% TFA over 120 min at a flow rate of 15 mL per min. Fractions were collected, analyzed by HPLC and fractions pooled to yield 11 mg.

Amino acid analysis: Arg 1.0 (1), Asx 1.09 (1), Glx 1.0 (1), Gly 1.06 (1), Leu 1.0 (1), Lys 1.87 (2), Phe 2.92 (3), Ser 2.29 (3), Val 2.04 (2).

FAB/MS: MH^+ 1787.5

EXAMPLE 5: Preparation of acetyl-tryptophyl-lysyl-valyl-arginyl-lysyl-seryl-phenylalanyl-phenylalanyl-lysyl-leucyl-glutamyl-glycyl-seryl-phenylalanyl-aspartyl-valyl-seryl-valine-amide (SEQ ID NO: 5)

The peptide was prepared on an ABI Model 431A Peptide Synthesizer using Version 1.12 of the standard BOC software. 4-methyl benzhydrylamine resin (0.58 g, 0.5 mmol) was used in the synthesis. The final weight of the resin was 2.26 g.

The peptide was cleaved from the resin (2.26 g) using 22 mL of HF and 2.2 mL of anisole for 60 min at 0°C. The resin was washed with ether and the peptide extracted with a 1:1 solution of TFA:CH_2Cl_2 to give 1.394 g of crude peptide.

The crude peptide (1.00 g) was purified on a Vydac C-18 column (15μ, 5 x 25 cm) eluting with a 30-55% gradient of 80% acetonitrile in 0.1% TFA over 120 min at a flow rate of 15 mL per min. Fractions were collected, analyzed by HPLC and pure fractions pooled and lyophilized to give 205 mg of semipure peptide.

Semipure peptide (205 mg) was purified using a Vydac C-18 column (15μ, 5 x 25 cm) eluting with a 25-50% gradient of 80% acetonitrile in 0.1% TFA over 120 min at a flow rate of 15 mL per min. Fractions were collected, analyzed and pooled to yield 83 mg.

Amino acid analysis: Arg 0.93 (1), Asx 1.05 (1), Glx 1.03 (1), Glx 1.04 (1), Leu 1.06 (1), Lys 2.81 (3), Phe 2.96 (3), Ser 2.28 (3), Trp 0.38 (1), Val 2.88 (3).

FAB/MS: MH^+ 2200.7

EXAMPLE 6: Preparation of D-arginyl-lysyl-seryl-phenylalanyl-phenylalanyl-lysyl-leucyl-glutamyl-glycyl-seryl-phenylalanyl-asparaginyl-valyl-seryl-valine-amide (SEQ ID NO: 6)

The peptide was prepared on an ABI Model 431A Peptide Synthesizer using Version 1.12 of the standard BOC software. 4-methyl benzhydrylamine resin (0.58 g, 0.5 mmol) was used in the synthesis. The final weight of the resin was 2.12 g.

The peptide was cleaved from the resin (2.06 g) using 20 mL of HF and 2.0 mL of anisole for 60 min at 0°C. The resin was washed with ether and the peptide extracted with a 1:1 solution of TFA:CH_2Cl_2 to give 1.18 g of crude peptide.

The crude peptide (900 mg) was purified on a Vydac C-18 column (15μ, 5 x 25 cm) eluting with a 25-50% gradient of 80% acetonitrile in 0.1% TFA over 120 min at a flow rate of 15 mL per min. Fractions were collected, analyzed by HPLC and pure fractions pooled and lyophilized to give 295 mg of semipure peptide.

Semipure peptide (295 mg) was purified on a Vydac C-18 column (10μ 2.2 x 25 cm) eluting with a 30-50% gradient of 80% acetonitrile in 0.1% TFA over 60 min at a flow rate of 10 mL per min. Fractions were collected, analyzed by HPLC and pooled to give 180 mg of pure product.

Amino acid analysis: Arg 0.96 (1), Asx 1.04 (1), Glx 0.99 (1), Gly 1.05 (1), Leu 1.01 (1), Lys 1.87 (2), Phe 2.85 (3), Ser 2.22 (3), Val 2.02 (2).

FAB/MS: MH^+ 1744.6

EXAMPLE 7: Preparation of D-tryptophyl-lysyl-valyl-arginyl-lysyl-seryl-phenylalanyl-phenylalanyl-lysyl-leucyl-glutamyl-glycyl-seryl-phenylalanyl-aspartic acid-amide (SEQ ID NO: 7)

The peptide was prepared on an ABI Model 431A Peptide Synthesizer using Version 1.12 of the standard BOC software. 4-methyl benzhydrylamine resin (0.58 g, 0.5 mmol) was used in the synthesis. The final weight of the resin was 2.03 g.

The peptide was cleaved from the resin (2.03 g) using 20 mL of HF and 2.0 mL of anisole for 60 min at 0°C. The resin was washed with ether and the peptide extracted with a 1:1 solution of TFA:CH_2Cl_2 to give 813 mg of crude peptide.

The crude peptide (813 mg) was purified on a Vydac C-18 column (15μ, 5 x 25 cm) eluting with a 15-45% gradient of 80% acetonitrile in 0.1% TFA over 120 min at a flow rate of 15 mL per min. Fractions were collected, analyzed by HPLC and pure fractions pooled and lyophilized to give 348 mg.

Amino acid analysis: Arg 0.94 (1), Asx 1.02 (1), Glx 1.0 (1), Gly 1.05 (1), Leu 1.01 (1), Lys 2.99 (3), Phe 2.99 (3), Ser 1.53 (2), Trp 0.39 (1), Val 1.01 (1).

FAB/MS: MH^+ 1875.5

EXAMPLE 8: Preparation of D-tryptophyl-lysyl-valyl-arginyl-lysyl-seryl-phenylalanyl-phenylalanyl-lysyl-leucyl-glutamyl-glycyl-seryl-phenylalanyl-aspartyl-valyl-seryl-valine-amide (SEQ ID NO: 8)

The peptide was prepared on an ABI Model 431A Peptide Synthesizer using Version 1.12 of the standard BOC software. 4-methyl benzhydrylamine resin (0.58 g, 0.5 mmol) was used in the synthesis. The final weight of the resin was 2.26 g.

The peptide was cleaved from the resin (2.15 g) using 21 mL of HF and 2.1 mL of anisole for 60 min at 0°C. The resin was washed with ether and the peptide extracted with a 1:1 solution of TFA:CH_2Cl_2 to give 1.31 g of crude peptide.

The crude peptide (1.00 g) was purified on a Vydac C-18 column (15μ, 5 x 25 cm) eluting with a 20-60% gradient of 80%acetonitrile in 0.1% TFA over 120 min at a flow rate of 15 mL per min. Fractions were collected, analyzed by HPLC and

pure fractions pooled and lyophilized to give 182 mg of semipure peptide.

Semipure peptide (182 mg) was purified on a Vydac C-18 column (10μ, 2.2 x 25 cm) eluting with a 30-50% gradient of 80% acetonitrile in 0.1% TFA over 60 min at a flow rate of 10 mL per min. Fractions were collected, analyzed by HPLC and pooled to give 88 mg of purified product.

Amino acid analysis: Arg 1.0 (1), Asx 1.01 (1), Glx 1.02 (1), Gly 1.0 (1), Leu 1.02 (1), Lys 2.96 (3), Phe 2.98 (3), Ser 2.26 (3), Trp 0.73 (1), Val 2.87 (3).

FAB/MS: MH^+ 2159.6

EXAMPLE 9:

The biological activity of the peptides prepared as described in Examples 1-8 was tested and determined as follows:

a. ELISA Assay to Determine Ability to Inhibit Binding of LPS to LBP

Using standard genetic engineering techniques, a fusion protein was assembled consisting of the entire mature human LBP coding region fused to the human IgG1 constant region. To allow secretion, the LBP-IgH fusion construct was co-expressed with a human kappa chain constant region (i.e., a light chain in which the entire variable region had been deleted) in murine myeloma cells. Functionally, then, the resulting LBP-immunoglobulin fusion protein, termed LBPfp, was designed such that the antigen binding V-region domains of an IgG molecule were replaced with the entire LBP molecule. Thus, the specificity of the LBP moiety for LPS was retained but was covalently linked to the effector functions of human IgG1 constant region domains.

To confirm the function of the LBPfp, an ELISA was carried out to measure the capacity of the LBBfp to bind LPS, and to be recognised by anti-human Fc antibodies. Different dilutions of protein A purified LBPfp were Fc-captured onto EIA plates previously coated with affinity purified anti-human Fc antibodies. After washing with PBS containing 1% low fat dry milk (PBSM), different dilutions of horseradish peroxidase (HRP) labelled

0111LPS (smooth) LPS were added to the wells and allowed to incubated for a further 30 minutes. After washing, the assay was developed by the addition of chromogenic substrate. Figure 1 shows the results of this assay. The LBPfp component appears to saturate at around 1μg/ml, but the LPS-HRP does not appear to saturate the Fc-captured LBPfp, even at 1/2500 dilution, the highest concentration tested. Nevertheless, since a strong signal was obtained using 1μg/ml LBPfp and 1/2500 LPS-HRP, these concentrations were selected as standard for subsequent screening assays.

The specificity of the ELISA is demonstrated in Fig 2. It can be seen that either solution-phase LBPfp or unlabelled 0111 LPS can competitively inhibit binding of LPS-HRP to fc-captured LBPfp, reaching 50% inhibition in the 1 μg/ml concentration range. Moreover, certain members of a panel of 26 different anti-human LBP monoclonal antibodies can also inhibit the binding of LPS-HRP to Fc-captured LBPfp (data not shown). Together, the results show that the Fc-captured LBPfp can specifically bind LPS-HRP in a way that can be inhibited by different specific blocking molecules, including monoclonal antibodies, unlabelled homologous LPS, or solution phase LBPfp.

Accordingly, the following standard competition ELISA-based screening assay was developed:

(1) in 50μl volumes, 1μg/ml of protein A-purified LBPfp was captured onto 96-microwell ELISA plates that had previously been coated with affinity purified anti-human Fc antibodies.

(2) after washing the assay wells with PBSM, a 1/2500 dilution of horseradish peroxidase-labeled 0111LPS (smooth LPS) was incubated with an equal volume (25μl)of competitor test peptide. The assay was allowed to incubate for 30 minutes, was washed three times, and developed by the addition of chromogen.

IC_{50} values for the peptides of Examples 1-8 were determined using this assay, and results are shown in Figure 3.

It is noteworthy that LPS-HRP is present in this ELISA assay at concentrations far exceeding (greater than 1000-fold) those typically found in patients with gram negative sepsis syndrome. Thus, the LPS neutralizing potency found in vitro for these peptides suggest that they and their analogs are attractive candidates for prevention or treatment of LPS toxicity in vivo.

b. Activity of peptides in complex biological milieux

It was of considerable interest to determine if the peptides of this invention could block complex physiological responses to LPS challenge. Two models were utilized to address this issue. First, the ability of the peptides to block the Limulus amoebocyte lysate (LAL) chromogenic reaction was tested (Biowhittaker kit# 50-648U). This assay depends on the ability of minute amounts of LPS (as little as 1pg/ml) to initiate a LAL endotoxin reaction. This cascade activates an enzyme which in turn releases p-nitroaniline from a synthetic substrate, a chromogenic reaction. Since the LAL endotoxin reaction is exqusitely sensitive to the presence of LPS, this format could provide a strigent test of the ability of the peptides to neutralize LPS challenge in a complex biological environment. Moreover, since the assay is sensitive to smooth and rough LPSs as well as lipid A, the assay presents an opportunity to determine whether the peptides are indeed specific for the lipid A moiety of LPS molecules.

The results of the LAL experiments are shown in Fig. 4. Fig. 4a compares the neutralization potency of the peptides of Ex. 1, 5, 4, and polymyxin B in a 0111LPS (smooth) challenge. These results show that the peptides tested can block smooth LPS challenge, but at approximately 0.2% the potency observed for polymyxin B. Figure 4b shows that the peptide of Example 1 can also inhibit the LAL reaction to J5 LPS, a rough form. In this case, polymyxin B appears to be even more potent, demonstrating a 2500-fold greater relative potency than the peptide of Example 1. Panel 4c compares the ability of the peptides to block

challenge of LAL with lipid A. In each case, the peptides could substantially inhibit the LAL reaction to lipid A. Hence, the peptides appear to neutralize both smooth and rough LPS as well as lipid A, and with similar potency in the IC50 10μM range. These results strongly suggest that the peptides of the invention are specific for the lipid A component of LPS.

A second model was utilized that, rather than emphasizing sensitivity, is perhaps more relevant to the clinical setting. In this model, human peripheral blood mononuclear cells (PBMC) are isolated from normal donors. These cells respond to LPS challenge by secreting a variety of cytokines such as IL-1, IL-6, and TNF alpha, which can be quantitated, providing an index of LPS stimulation. Moreover, PBMC sensitivity to LPS is potentiated by as much as 1000-fold by the addition of LBP, either in pure form, or as a constituent of added serum, responding to as little as 10 pg/ml LPS in the presence of 1μg/ml LBP. Figure 5 shows the ability of the peptide of Example 1 to block PBMC response to LPS challenge in serum free, purified LBP-potentiated cultures. The results show that in the absence of the peptide, PBMC cultures respond maximally to stimulation with about 1ng/ml LPS. As little as 3μM of the peptide can completely block the TNF response to this maximal LPS stimulation. It is interesting that the peptide mediated LPS neutralization could be overwhelmed by the addition of increased LPS doses, in a roughly stoichiometric fashion. This observation suggests that the ability of the peptide to neutralize the PBMC LPS response reflects its LPS binding properties, and in particular does not reflect toxicity to the PBMC. In this regard, in separate experiments the effect of the peptide on viability of several different cell types including PBMC was tested, and was found to have no measurable effect, even at a 5-fold greater concentration than the maximum used here (not shown). Thus, the results show that, in vitro, the peptide can completely inhibit the normal human PBMC TNF response to LPS (up to 10 ng/ml LPS in

this experiment) and which, notably, are substantially greater LPS levels than ypically found in the clinical setting.

The peptide of Example 6 is otherwise identical to that of Example 1, but has the normal n-terminal residue L-Arg replaced with a D-Arg residue. This analog was tested in the PBMC format for the ability to neutralize LPS in the presence of normal human serum. Figure 6 shows the result of one such experiment in which the peptide of Example 6 was tested for LPS neutralization potency in the presence of 1% normal human serum. The results show that this peptide can completely inhibit stimulation with 1ng/ml LPS, with IC50 values of less than 10μM. While this potency is somewhat less than found for the peptide of Example 1 in the serum free, purified LBP potentiated format, it nevertheless demonstrates a large degree of serum stability. Thus, the results indicate the peptides of this invention retain LPS neutralization properties when tested in complex biological milieux, including normal human serum. Even more importantly, these peptides can completely inhibit cellular recognition of LPS in vitro, abrogating cytokine secretion, known to play a central role in the pathology of gram negative sepsis syndrome.

SEQUENCE LISTING

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(2) INFORMATION FOR SEQ ID NO:1:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:1:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:2:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 18 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:2:

Trp Lys Val Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp
1 5 10 15

Val Ser Val

(2) INFORMATION FOR SEQ ID NO:3:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:3:

Trp Lys Val Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:4:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(ix) FEATURE:
(A) NAME/KEY: Modified-site
(B) LOCATION: 1
(D) OTHER INFORMATION: Acetylated

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:4:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:5:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 18 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(ix) FEATURE:
(A) NAME/KEY: Modified-site
(B) LOCATION: 1
(D) OTHER INFORMATION: Acetylated

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:5:

Trp Lys Val Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp
1 5 10 15

Val Ser Val

(2) INFORMATION FOR SEQ ID NO:6:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:6:

Gln Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:7:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:7:

Lys Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:8:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:8:

Ala Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:9:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:9:

His Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:10:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:10:

Arg Arg Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:11:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues

(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:11:

Arg Ala Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:12:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:12:

Arg His Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:13:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:13:

Arg Lys Thr Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:14:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:14:

Arg Lys Ala Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:15:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:15:

Arg Lys Arg Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:16:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:16:

Arg Lys Ser Ala Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:17:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:17:

Arg Lys Ser Tyr Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:18:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues

(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:18:

Arg Lys Ser Ser Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:19:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:19:

Arg Lys Ser Pro Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:20:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:20:

Arg Lys Ser Phe Tyr Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:21:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:21:

Arg Lys Ser Phe Leu Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:22:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:22:

Arg Lys Ser Phe Ala Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:23:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:23:

Arg Lys Ser Phe Phe Arg Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:24:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:24:

Arg Lys Ser Phe Phe Ala Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:25:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues

(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:25:

Arg Lys Ser Phe Phe His Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:26:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:26:

Arg Lys Ser Phe Phe Lys Ala Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:27:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:27:

Arg Lys Ser Phe Phe Lys Met Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:28:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:28:

Arg Lys Ser Phe Phe Lys Leu Asn Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:29:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:29:

Arg Lys Ser Phe Phe Lys Leu Lys Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:30:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:30:

Arg Lys Ser Phe Phe Lys Leu Ser Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:31:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:31:

Arg Lys Ser Phe Phe Lys Leu Ala Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:32:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues

(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:32:

Arg Lys Ser Phe Phe Lys Leu Gln Ala Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:33:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:33:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Thr Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:34:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:34:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Asn Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:35:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:35:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ala Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:36:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:36:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Tyr Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:37:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:37:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Ala Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:38:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:38:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Glu Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:39:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues

(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:39:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Ala Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:40:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:40:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe His Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:41:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:41:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Lys Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:42:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:42:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Arg Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:43:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:43:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Ala Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:44:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:44:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Leu Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:45:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:45:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Lys Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:46:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues

(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:46:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ala Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:47:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:47:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Thr Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:48:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:48:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Tyr Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:49:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:49:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Tyr
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:50:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:50:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Ala
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:51:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:51:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Ile
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:52:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 17 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:52:

Lys Val Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val
1 5 10 15

Ser Val

(2) INFORMATION FOR SEQ ID NO:53:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 16 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:53:

Val Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser
1 5 10 15

Val

(2) INFORMATION FOR SEQ ID NO:54:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:54:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:55:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 14 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:55:

Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser Val
1 5 10

(2) INFORMATION FOR SEQ ID NO:56:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 17 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:56:

Trp Lys Val Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp
1 5 10 15

Val Ser

(2) INFORMATION FOR SEQ ID NO:57:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 16 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:57:

Trp Lys Val Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp
1 5 10 15

Val

(2) INFORMATION FOR SEQ ID NO:58:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 15 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:58:

Trp Lys Val Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp
1 5 10 15

(2) INFORMATION FOR SEQ ID NO:59:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 14 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:59:

Trp Lys Val Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe
1 5 10

(2) INFORMATION FOR SEQ ID NO:60:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 13 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:60:

Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser
1 5 10

(2) INFORMATION FOR SEQ ID NO:61:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 14 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:61:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp Val Ser
1 5 10

(2) INFORMATION FOR SEQ ID NO:62:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 14 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:62:

Lys Val Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp
1 5 10

(2) INFORMATION FOR SEQ ID NO:63:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 12 amino acid residues

(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:63:

Arg Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp
1 5 10

(2) INFORMATION FOR SEQ ID NO:64:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 11 amino acid residues
(B) TYPE: Amino Acid
(C) STRANDEDNESS:
(D) TOPOLOGY: Linear

(ii) MOLECULE TYPE: Peptide

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:64:

Lys Ser Phe Phe Lys Leu Gln Gly Ser Phe Asp
1 5 10

WHAT IS CLAIMED IS:

1. A peptide capable including a sequence consisting essentially of the Formula:

R^1-X-A-B-C-D-E-F-G-H-I-J-Y-R^2 wherein:

A is selected from the group consisting of D- or L-lysine, D- or L-arginine, D- or L-alanine, and D- or L-hystidine;

B is selected from the group consisting of D- or L-serine, D- or L-threonine, D- or L-alanine, and D- or L-arginine;

C is selected from the group consisting of D- or L-phenylalanine, D- or L-alanine, D- or L-tyrosine, D- or L-serine, and D- or L-proline;

D is selected from the group consisting of D- or L-phenylalanine, D- or L-tyrosine, D- or L-leucine, and D- or L-alanine;

E is selected from the group consisting of D- or L-lysine, D- or L-arginine, D- or L-alanine, and D- or L-histidine;

F is selected from the group consisting of D- or L-leucine, D- or L-alanine, and D- or L-methionine;

G is selected from the group consisting of D- or L-glutamine, D- or L-asparagine, D- or L-lysine, D- or L-serine, and D- or L-alanine;

H is selected from the group consisting of D- or L-glycine, and D- or L-alanine;

I is selected from the group consisting of D- or L-serine, D- or L-threonine, D- or L-asparagine; and D- or L-alanine;

J is selected from the group consisting of D- or L-phenylalanine; D- or L-alanine, and D- or L-tyrosine;

X is a linear chain of from zero to four amino acids;

Y is a linear chain of from zero to five amino acids;

R^1 is H (signifying a free-terminal amino group), formyl, lower alkyl, aryl, lower alkanoyl, aroyl, biotinyl,

alkyloxycarbonyl, aryloxycarbonyl or desamino (signifying no alpha amino group on the N-terminal amino acid); and

R^2 is H (signifying descarboxy where the α carboxyl group in the C-terminal amino acid is absent) or OR^3, where R^3 is H (signifying a free carboxylic acid on a C-terminal carboxyl group) or lower alkyl or aryl, NR^4 R^5 where R^4 and R^5 are each selected independently from H, lower alkyl, aryl, or taken together are a methylene chain of 4-8 methylene groups $(-(CH_2)_n-$ where n = 4 to 8);

or a pharmaceutically acceptable acid- or base-addition salt thereof.

2. A peptide of Claim 1 wherein X is selected from the group consisting of:

Trp-Lys-Val-Arg
Lys-Val-Arg
Val-Arg
Arg
Gln
Lys
Ala
His
and null (signifying no amino acid)

or analogs thereof wherein one or more of the amino acids are replaced with the analogous D-amino acid.

3. A peptide of Claim 1 wherein Y is selected from a group consisting of

Asp
Asp-Val-Ser-Val
Asp-Val
Asp-Val-Ser
Glu-Val-Ser-Val
Ala-Val-Ser-Val
His-Val-Ser-Val
Lys-Val-Ser-Val
Arg-Val-Ser-Val

Asp-Val-Ser-Val
Asp-Leu-Ser-Val
Asp-Lys-Ser-Val
Asp-Val-Ala-Val
Asp-Val-Thr-Val
Asp-Val-Tyr-Val
Asp-Val-Ser-Val
Asp-Val-Ser-Ala
Asp-Val-Ser-Ile
and null (signifying no amino acid);
or analogs thereof wherein one or more of the amino acids are replaced with the analogous D-amino acid.

4. A biologically active peptide of Claim 1 selected from the group consisting of:

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 1)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 2)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2 (SEQ ID NO: 3)

Acetyl-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 4)

Acetyl-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 5)

D-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2

D-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2

D-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2

Gln-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 6)

Lys-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 7)

Ala-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 8)

His-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 9)

Arg-Arg-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 10)

Arg-Ala-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 11)

Arg-His-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 12)

Arg-Lys-Thr-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 13)

Arg-Lys-Ala-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 14)

Arg-Lys-Arg-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 15)

Arg-Lys-Ser-Ala-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 16)

Arg-Lys-Ser-Tyr-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 17)

Arg-Lys-Ser-Ser-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 18)

Arg-Lys-Ser-Pro-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 19)

Arg-Lys-Ser-Phe-Tyr-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 20)

Arg-Lys-Ser-Phe-Leu-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 21)

Arg-Lys-Ser-Phe-Ala-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 22)

Arg-Lys-Ser-Phe-Phe-Arg-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 23)

Arg-Lys-Ser-Phe-Phe-Ala-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 24)

Arg-Lys-Ser-Phe-Phe-His-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 25)

Arg-Lys-Ser-Phe-Phe-Lys-Ala-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 26)

Arg-Lys-Ser-Phe-Phe-Lys-Met-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 27)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Asn-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 28)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Lys-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 29)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Ser-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 30)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Ala-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 31)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Ala-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 32)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Thr-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 33)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Asn-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 34)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ala-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 35)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Tyr-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 36)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Ala-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 37)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Glu-Val-Ser-Val-NH_2 (SEQ ID NO: 38)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Ala-Val-Ser-Val-NH_2 (SEQ ID NO: 39)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-His-Val-Ser-Val-NH_2 (SEQ ID NO: 40)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Lys-Val-Ser-Val-NH_2 (SEQ ID NO: 41)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Arg-Val-Ser-Val-NH_2 (SEQ ID NO: 42)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Ala-Ser-Val-NH_2 (SEQ ID NO: 43)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Leu-Ser-Val-NH_2 (SEQ ID NO: 44)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Lys-Ser-Val-NH_2 (SEQ ID NO: 45)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ala-Val-NH_2 (SEQ ID NO: 46)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Thr-Val-NH_2 (SEQ ID NO: 47)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Tyr-Val-NH_2 (SEQ ID NO: 48)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Tyr-NH_2 (SEQ ID NO: 49)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Ala-NH_2 (SEQ ID NO: 50)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Ile-NH_2 (SEQ ID NO: 51)

Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 52)

Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 53)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 54)

Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 55)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-NH_2 (SEQ ID NO: 56)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-NH_2 (SEQ ID NO: 57)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2 (SEQ ID NO: 58)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-NH_2 (SEQ ID NO: 59)

Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-NH_2 (SEQ ID NO: 60)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-NH_2 (SEQ ID NO: 61)

Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2 (SEQ ID NO: 62)

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2 (SEQ ID NO: 63)

Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2 (SEQ ID NO: 64)

D-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-Lys(Biotin)-NH_2

D-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2.

5. A peptide of Claim 2 where Y is selected from the group consisting of

Asp
Asp-Val-Ser-Val
Asp-Val
Asp-Val-Ser
Glu-Val-Ser-Val
Ala-Val-Ser-Val
His-Val-Ser-Val
Lys-Val-Ser-Val
Arg-Val-Ser-Val
Asp-Val-Ser-Val
Asp-Leu-Ser-Val
Asp-Lys-Ser-Val
Asp-Val-Ala-Val
Asp-Val-Thr-Val
Asp-Val-Tyr-Val
Asp-Val-Ser-Val
Asp-Val-Ser-Ala
Asp-Val-Ser-Ile
and null (signifying no amino acid);

or analogs thereof wherein one or more of the amino acids are replaced with the analogous D-amino acid.

6. A peptide of Claim 4 selected from the group consisting of

Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 1)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 2)

Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2 (SEQ ID NO: 3)

Acetyl-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 4)

Acetyl-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2 (SEQ ID NO: 5)

D-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2

D-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-NH_2

D-Trp-Lys-Val-Arg-Lys-Ser-Phe-Phe-Lys-Leu-Gln-Gly-Ser-Phe-Asp-Val-Ser-Val-NH_2

7. A pharmaceutical composition comprising a biologically active peptide of claim 1 in an amount effective to inhibit LPS and a pharmaceutically acceptable carrier or diluent.

8. The pharmaceutical composition of claim 7 wherein said pharmaceutically acceptable carrier or diluent is acceptable for parental administration.

9. The pharmaceutical composition of claim 7 wherein said pharmaceutically acceptable carrier or diluent is acceptable for oral administration.

10. A method of inhibiting LPS in a host comprising the step of administering to said host a biologically active peptide of claim 1 in an amount effective to inhibit binding of LPS to LBP.

11. A method of detecting LPS or LPS fragments in a host comprising the steps of:

(a) taking a blood or tissue sample to be tested from said host;

(b) contacting said sample to be tested with a labeled peptide of Claim 1; and

(c) assessing the binding of said labeled peptide to components of said sample to be tested.

12. The method of Claim 11 wherein said peptide is labeled with a moiety selected from the group consisting of radioactive tracers, fluorescent tags, enzymes, and electron-dense materials.

13. A method of preparing a peptide of Claim 1 comprising adding amino acids either singly or in preformed blocks of amino acids to an appropriately functionalized solid support.

14. A method of Claim 1 wherein said amino acids are assembled either singly or in preformed blocks in solution or suspension by chemical ligation techniques.

15. A method of preparing a peptide of Claim 1 wherein said amino acids are assembled either singly of in preformed blocks in solution or suspension by enzymatic ligation techniques.

16. A method of preparing a peptide of Claim 1 wherein said peptide is produced enzymatically by inserting nucleic acids, coding the peptide into an expression vector, expressing the DNA, translating the DNA into RNA and translating the RNA into the peptide.

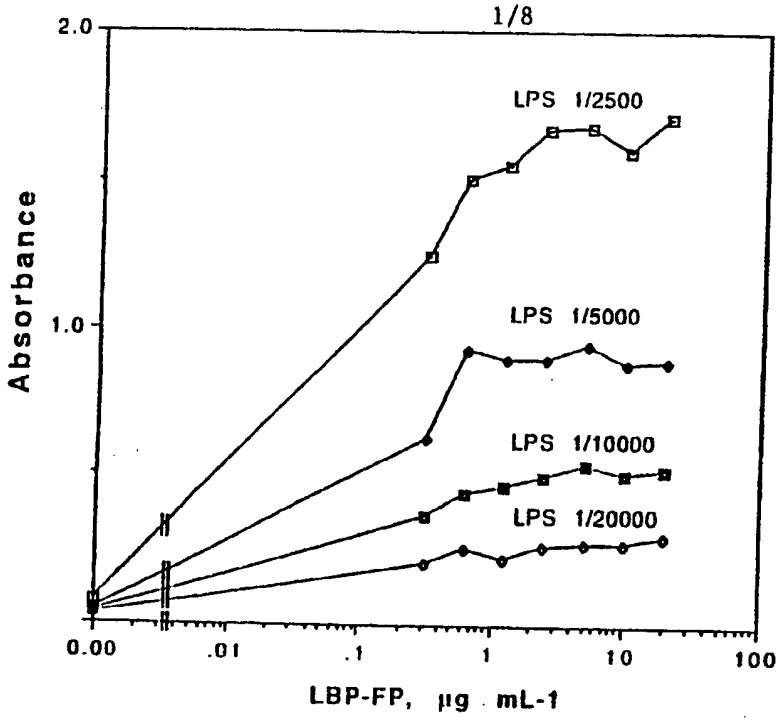
2.0
1.0
Absorbance
LPS 1/2500
LPS 1/5000
LPS 1/10000
LPS 1/20000
0.00
.01
.1
1
10
100
LBP-FP, µg mL-1

FIGURE 1

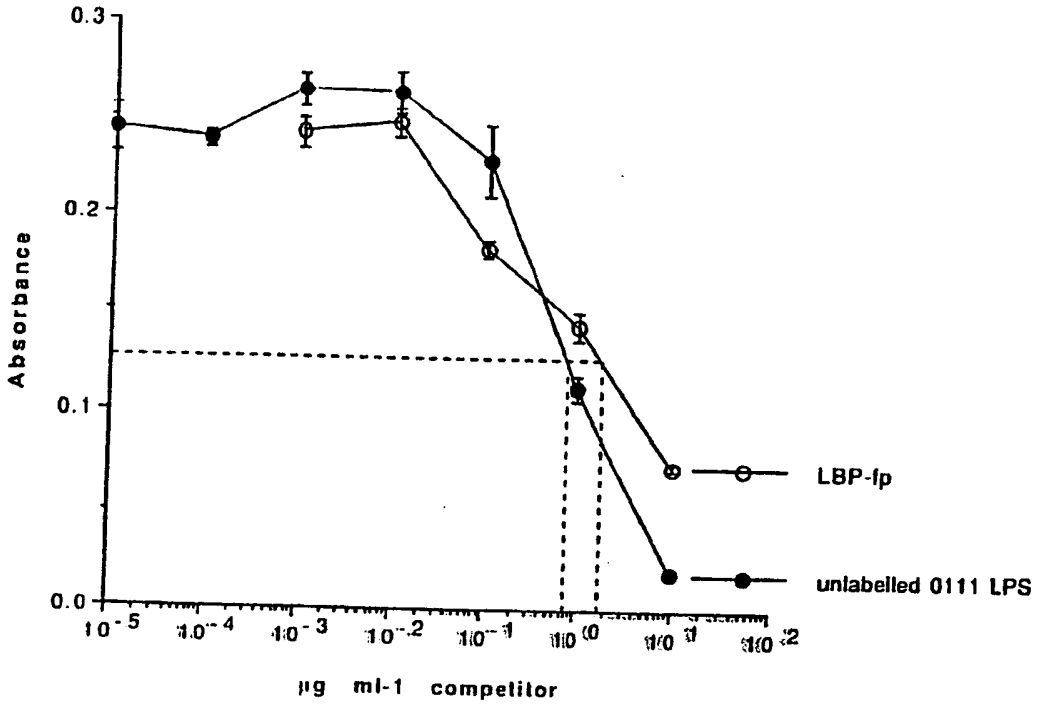
0.3
0.2
0.1
0.0
Absorbance
LBP-fp
unlabelled 0111 LPS
µg ml-1 competitor

FIGURE 2

IC50 of Peptides of Examples 1-8
(ELISA competitive inhibition assay)

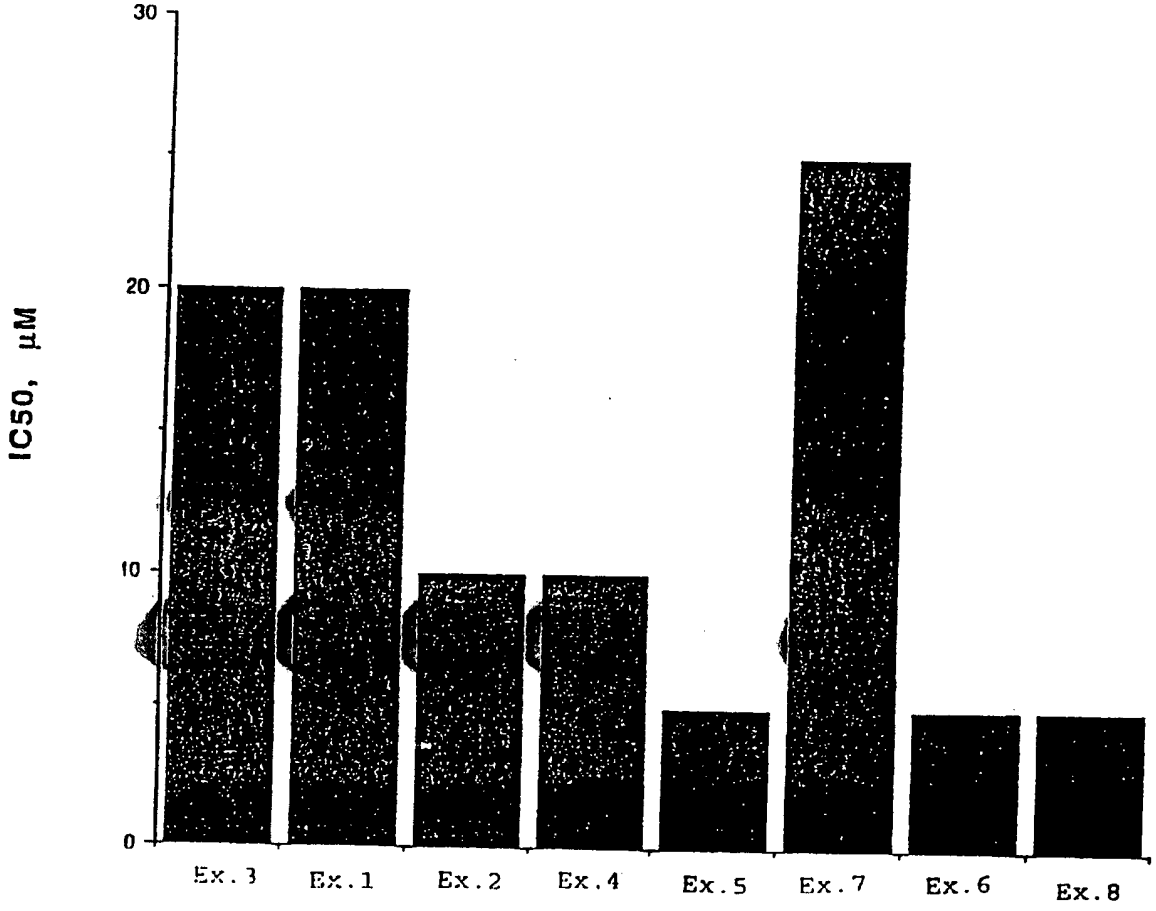

FIGURE 3

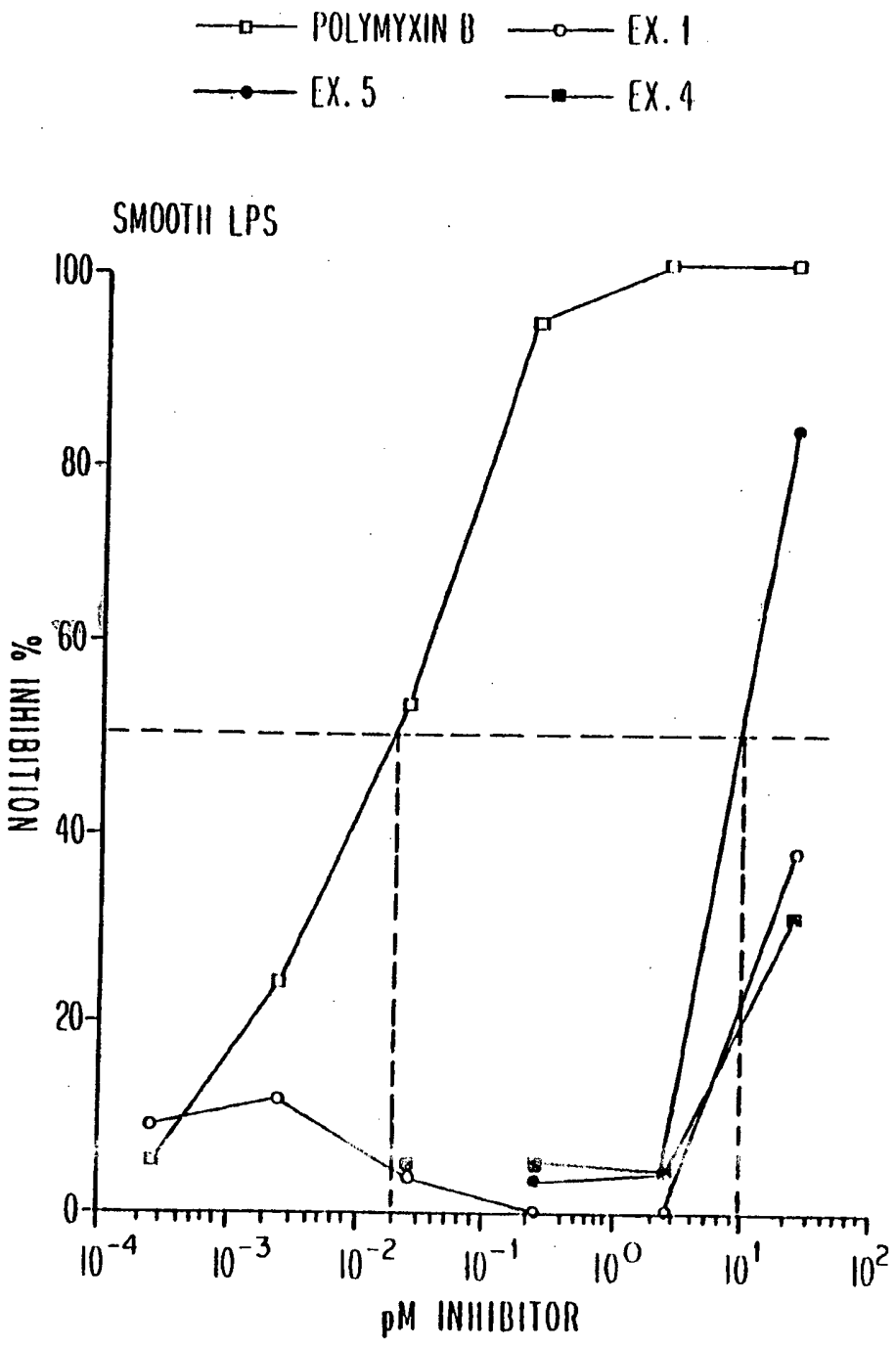
POLYMYXIN B
EX. 1
EX. 5
EX. 4
SMOOTH LPS
100
80
60
40
20
0
% INHIBITION
10^{-4}
10^{-3}
10^{-2}
10^{-1}
10^{0}
10^{1}
10^{2}
pM INHIBITOR

Fig. 4A

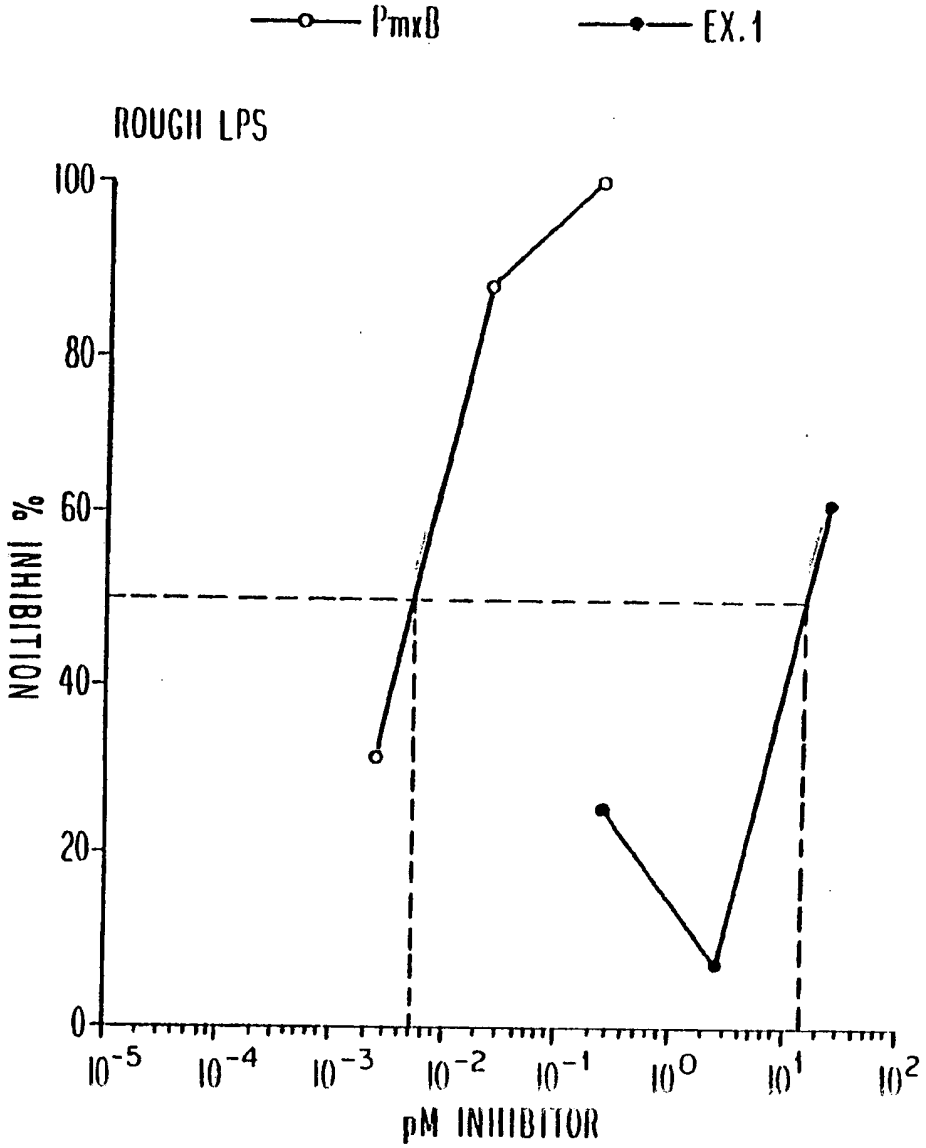
PmxB
EX.1
ROUGH LPS
100
80
60
40
20
0
% INHIBITION
10^{-5}
10^{-4}
10^{-3}
10^{-2}
10^{-1}
10^{0}
10^{1}
10^{2}
pM INHIBITOR

Fig. 4B

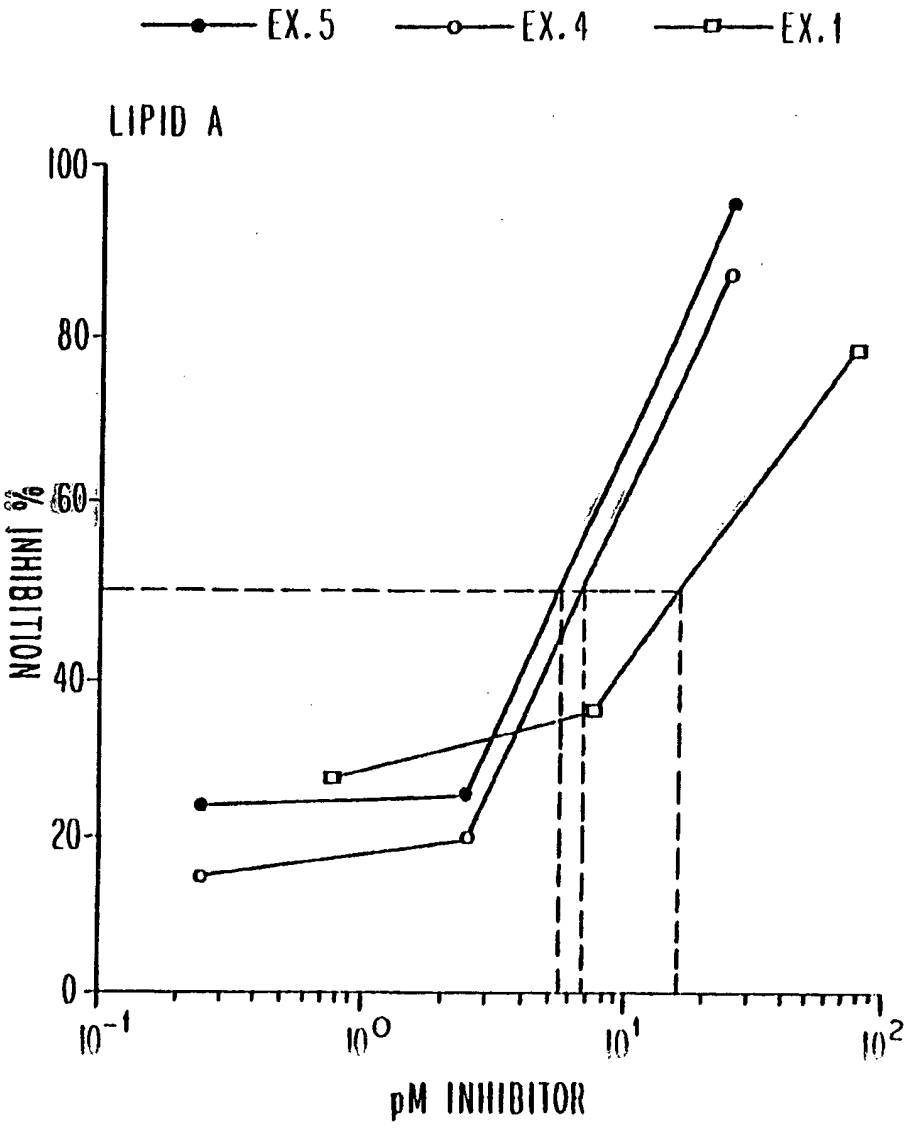
EX.5
EX.4
EX.1
LIPID A
100
80
60
40
20
0
% INHIBITION
10^{-1}
10^{0}
10^{1}
10^{2}
pM INHIBITOR

Fig. 4C

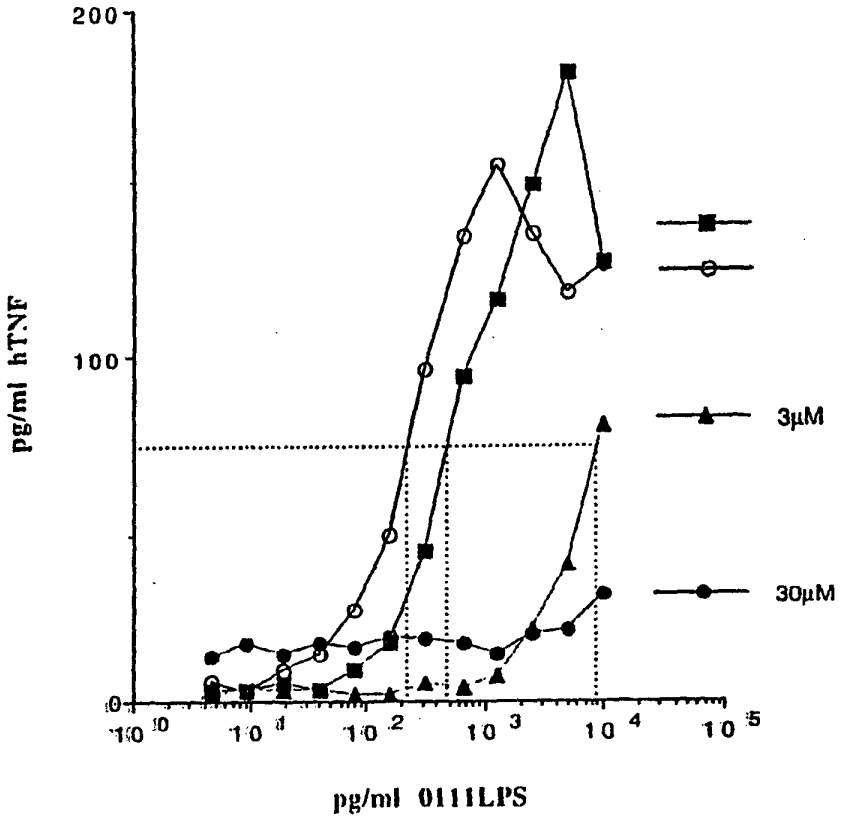
200
100
0
pg/ml hTNF
3μM
30μM
10 10
10 1
10 2
10 3
10 4
10 5
pg/ml 0111LPS

FIGURE 5

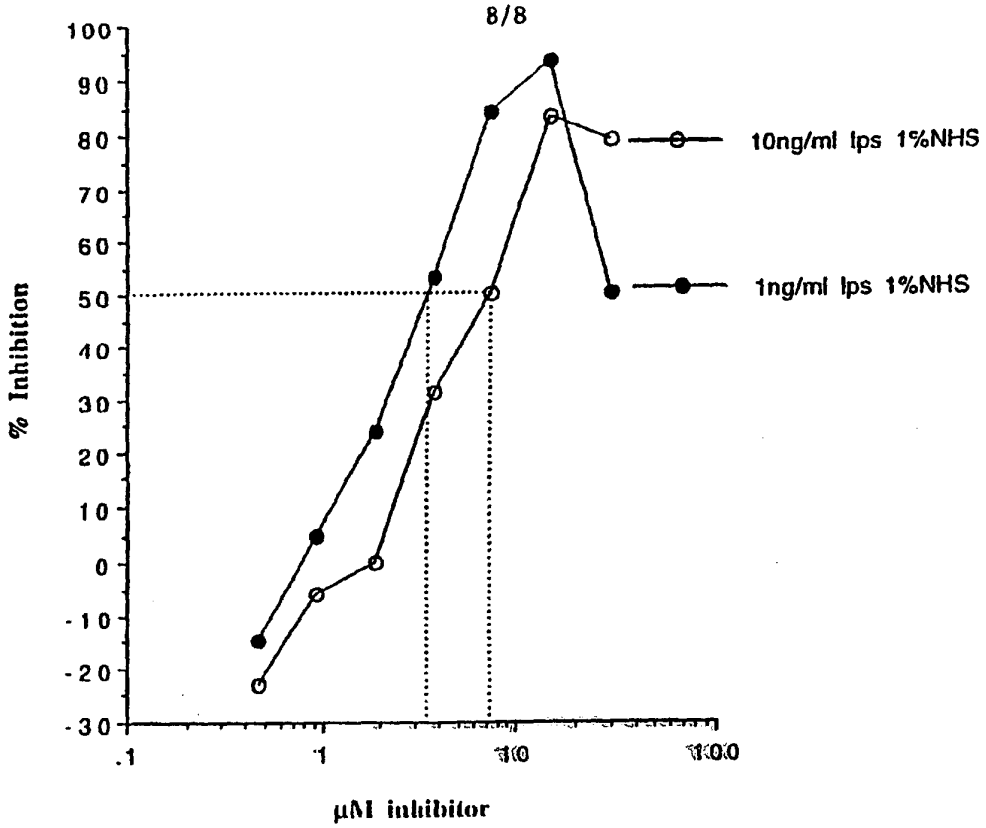
100
90
80
70
60
50
40
30
20
10
0
-10
-20
-30
% Inhibition
.1
1
10
100
µM inhibitor
10ng/ml lps 1%NHS
1ng/ml lps 1%NHS

FIGURE 6

INTERNATIONAL SEARCH REPORT

Int. ..ational application No.
PCT/US94/10760

A. CLASSIFICATION OF SUBJECT MATTER

IPC(6) :Please See Extra Sheet.
US CL :530/326, 327, 328, 333, 334, 338; 514/13,14,15; 435/68.1, 69.1; 536/25.3
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 530/326, 327, 328, 333, 334, 338; 514/13,14,15; 435/68.1, 69.1; 536/25.3

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

APS, DIALOG, SEQUENCE SEARCH DATA BASES, search terms: sequence, lipopolysaccharide binding protein, LBP, domain

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	The EMBO Journal, Volume 12, Number 9, issued 01 September 1993, Hoess et al. "Crystal structures of an endotoxin-neutralizing protein from the horseshoe crab, *Limulus* anti-LPS factor, at 1.5 A resolution", pages 3351-3356, see the whole publication, especially the abstract and Figure 1.	1-16
Y	Analytical Biochemistry, Volume 197, issued 1991, Valerio et al., "Synthesis of peptide analogues using the multipin peptide synthesis method", pages 168-177, see whole publication.	1-16
Y	US, A, 5,334,564 (SCOTT ET AL.) 02 August 1994, col. 5, lines 10-14 and cols. 11 and 12.	1-16

[X] Further documents are listed in the continuation of Box C. [ ] See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
14 DECEMBER 1994	10 JAN 1995

Name and mailing address of the ISA/US	Authorized officer
Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231	KEITH FURMAN, PH.D.
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Form PCT/ISA/210 (second sheet)(July 1992)*

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US94/10760

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	Science, Volume 249, issued 21 September 1990, Schumann et al. "Structure and function of lipopolysaccharide binding protein", pages 1429-1433, see Figure 1.	1-16
A,P	Bio/Technology, Volume 12, issued August 1994, Highfield, "Sepsis: the more the murkier", page 828, see page 828.	7-10
A	Archives of Surgery, Volume 128, issued January 1993, Geller et al., "Induction of hepatocyte lipopolysaccharide binding protein in models of sepsis and the acute-phase response", pages 22-28, see the whole publication, especially the abstract.	7-10
A,P	The Journal of Immunology, Volume 151, Number 7, issued 01 October 1993, Gessani et al., "Enhanced production of LPS-induced cytokines during differentiation of human monocytes to macrophages", pages 3758-3766, see column 2 of page 3765.	7-10
A,P	Proceedings of the National Academy of Sciences USA, Volume 90, issued November 1993, Gallay et al., "Lipopolysaccharide-binding protein as a major plasma protein responsible for endotoxemic shock", pages 9935-9938, see column 1 of page 9935.	7-10
A	Infection and Immunity, Volume 61, Number 7, issued July 1993, Cross et al., "Choice of bacteria in animal models of sepsis", pages 2741-2747, see whole publication, especially page 2741.	1-16

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US94/10760

A. CLASSIFICATION OF SUBJECT MATTER:
IPC (6):

C07K 1/02, 1/04, 1/06, 1/08, 1/10, 1/107, 1/113, 7/04, 7/06, 7/08, 14/47; C12N 15/10; A61K 38/08, 38/10, 38/17; G01N 33/53, 33/58, 33/60, 33/68, 33/567